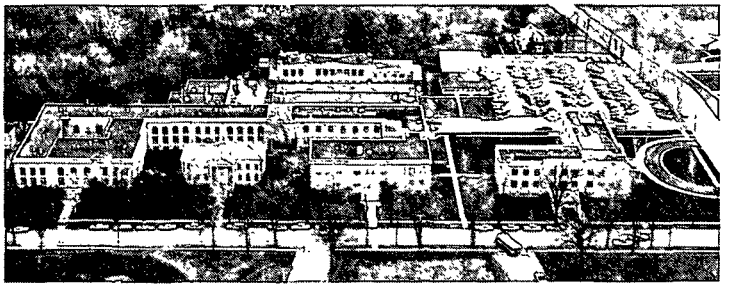

THE INSTITUTE OF PAPER CHEMISTRY, APPLETON, WISCONSIN

IPC TECHNICAL PAPER SERIES
NUMBER 224

THE STRUCTURES OF CELLULOSE

RAJAI H. ATALLA

FEBRUARY, 1987

The Structures of Cellulose

Rajai H. Atalla

This manuscript is based on results obtained in student research and dues funded research, and has been submitted for inclusion in the Symposium on Solid State Characterization of Cellulose, R. H. Atalla, Editor, to be published in the ACS Symposium Series

The Structures of Cellulose

Rajai H. Atalla

The Institute of Paper Chemistry
Appleton, Wisconsin 54912

An overview of studies of the structure of cellulose is presented and begins with a historical perspective, developed with particular emphasis on the early diffractometric studies. More recent studies are then described, and the key questions confronted in any analysis of diffractometric data are discussed. The central questions are concerned with the validity of the assumption that the unit cells of cellulose belong to space group $P2_1$, and whether the twofold screw axis associated with this space group coincides with the molecular chain axes. The diversity of the interpretations which occur in the literature and in following chapters is noted. More recent spectroscopic investigations are then discussed, with emphasis on the degree to which they may provide additional information concerning structure. It is noted that although both Raman spectroscopy and CP-MAS ^{13}C NMR cannot provide direct information concerning the positions of molecules in the unit cells, they are sensitive to the values of the internal coordinates. Thus, they provide information complementary to the diffractometric data in that it serves to constrain the acceptable structural models to a smaller subset than that otherwise admissible on the basis of diffractometric observations alone. In this respect, the spectroscopic information complements the diffractometric data in the same way as the assumptions concerning the symmetry of the unit cell. Furthermore, it appears that the structures suggested by the spectroscopic studies represent relatively small although significant departures from those derived on the basis of diffractometry alone. In anticipation of future directions in studies of celluloses, it is noted that multidisciplinary approaches, similar to some described in later chapters, hold great promise for future progress in understanding the structural diversity that is characteristic of cellulose.

Since the occurrence of cellulose as a distinct substance was first recognized by Anselme Payen in 1842 the evolution of ideas concerning its structure has been closely related to advances in structural chemistry and its methodologies. The pattern of close relation continues into the present time and is well reflected in the following chapters which include contributions from most of the major laboratories active in the field. In this chapter, we discuss the structural problem in general and place those of the following chapters which are concerned with the problem in perspective relative to recent developments in the field, with particular emphasis on the past decade.

The procedures for structural studies on cellulose have much in common with investigations of structure in polymers in general. In most instances diffractometric data are not sufficient for a solution of the structure in a manner analogous to that possible for lower molecular weight compounds which can be made to form single crystals. It becomes necessary, therefore, to complement diffractometric data with structural information derived from studies carried out on the monomers or oligomers.

Kakudo and Kasai have summarized the central problem well (1): "There are generally less than 100 independently observable diffractions for all layer lines in the x-ray diagram of a fibrous polymer. This clearly imposes limitations on the precision which can be achieved in polymer structure analysis, especially in comparison with the 2000 or more diffractions observable for ordinary single crystals. However, the molecular chains of the high polymer usually possess some symmetry of their own, and it is often possible to devise a structural model of the molecular chain to interpret the fiber period in terms of the chemical composition by comparison with similar or homologous substances of known structure. Structural information from methods other than x-ray diffraction (e.g., infrared and NMR spectroscopy) are also sometimes helpful in devising a structural model of the molecular chain. The majority of the structural analyses which have so far been performed are based on models derived in this way. This is, of course, a trial and error method". Similar perspectives have been presented by Arnott (2), Atkins (3), and Tadokoro (4,5).

An acceptable fit to the diffractometric data is not the ultimate objective, however. Rather it is the development of a model that possesses a significant measure of validity as the basis for organization, explanation and prediction of experimental observations. With respect to this criterion, the models of cellulose which have been developed so far leave much to be desired, for their capacity to integrate and unify the vast array of information concerning cellulose is limited indeed. One of the objectives of this symposium is to facilitate identification of points of departure for further studies in search of models which are more useful.

To help place the proceedings in perspective we begin with a brief historical review, and continue with a discussion of recent contributions based on the key methodologies which have been used. The methodologies are in three broad, complementary categories, which include diffractometry, spectroscopy, and theoretical model building on the basis of conformational analysis. Although, significant structural information is inferred from patterns of chemical

reactions under a wide range of conditions, we limit this chapter to studies based on physical methods.

In order to achieve greater clarity in the following discussion, it is well to note that questions of structure arise at three different levels. The first, that of the chemical structure, reflects the pattern of covalent bonding in cellulose molecules and is generally well established. While the evolution of concepts at this level is of historical interest, it is not under discussion in these proceedings. The next level of structure is that of the relative organization of the repeat units in an individual molecule, under constraints of conformational energy considerations, as well as considerations of packing of the molecules in a particular state of aggregation. This level of structure is particularly important in spectroscopic studies wherein the energy levels between which transitions are observed are determined by the values of the internal coordinates which define molecular conformations. The final level of structure is that reflecting the arrangement of the molecules relative to each other in a particular state of aggregation, whether it be amorphous, or represents one or another of the crystalline allomorphs which occur because of the polymorphy characteristic of the crystallinity of cellulose. This is the level of structure probed by diffractometric measurements which are inherently most sensitive to the three dimensional organization represented by a particular state of aggregation.

Historical Overview

The evolution of ideas concerning the nature of cellulose and the models of its chemical structure have been described by Purves (6) in an excellent overview, beginning with the first observations by Payen and leading up to those which finally won acceptance of the polymer hypothesis in the decade immediately preceding the Second World War. Another valuable perspective is presented by Flory (7) in his general review of the evolution of the polymeric hypothesis, highlighting investigations of the three common natural homopolymers: starch, cellulose, and natural rubber. Finally, the first chapter in the treatise by Hermans (8) focuses on the physical chemical aspects of the early structural studies, in an account which is an excellent complement to the review by Purves with its emphasis on the classical organic chemical phase in the structural studies.

Among more recent reviews of structure, those by Jones (9), and by Tonessen and Ellefsen (10,11) are the most comprehensive. Preston (12) and Frey-Wyssling (13) in their respective treatises on plant cell walls, have also touched upon the problem of the structure of cellulose. The reader is referred to these sources for comprehensive presentations of the range of proposals concerning the structures of cellulose which have been under discussion in recent decades. A representative subset will be presented here as a point of departure for following discussions.

Quite early in the x-ray diffractometric studies of cellulose it was recognized that its crystallinity is polymorphic. It was established that native cellulose, on the one hand, and both regenerated and mercerized celluloses, on the other, represent two distinct crystallographic allomorphs (14). Little has transpired

since the early studies to change these perceptions. There has been, however, little agreement regarding the structures of the two forms. For example, Petitpas et al. (15) have suggested on the basis of extensive analyses of electron-density distributions from x-ray diffractometric measurements that chain conformations are different in celluloses I and II. In contrast, Norman (16) has interpreted the results of his equally comprehensive x-ray diffractometric studies in terms of similar conformations for the two allomorphs.

At a more basic level than the comparison of celluloses I and II, the stucture of the native form itself has remained in question. Among recent studies, for example, Blackwell and Gardner (17), in their analysis of the structure of cellulose from Valonia ventricosa, assumed a lattice belonging to the $P2_1$ space group, with the twofold screw axis coincident with the molecular chain axis. Hebert and Muller (18), on the other hand, in an electron diffractometric study of a number of celluloses including Valonia, confirmed the findings of earlier investigators who found no systematic absences of the odd order reflec-tions forbidden by the selection rules of $P2_1$, and concluded that the cellulose unit cells do not belong to that space group.

Even when $P2_1$ is taken to be the appropriate space group, the question of chain polarity remains. As noted by Jones (19), and by Howsmon and Sisson (20), the structure initially proposed by Meyer and Mark (21) assumed that the chains were parallel in polarity. The structure later proposed by Meyer and Misch (22) was based on the reasoning that the rapidity of mercerization, and its occurrence without dissolution required that the polarity of the chains be the same in both celluloses I and II. It was reasoned further that regeneration of cellulose from solution is most likely to result in precipitation in an antiparallel form, and that the similarity between x-ray diffraction patterns of mercerized and regenerated cellulose required that they have the same polarity. It was thus inferred that native cellulose must also have an antiparallel structure.

Although the argument that regeneration in the antiparallel mode is more probable was found invalid within a decade of its first presentation (23), the relative organization of molecules suggested by Meyer and Misch remained the point of departure for most subsequent investigators.

When the models incorporating antiparallel arrangement of the chains are extended to native cellulose, they pose serious questions concerning proposed mechanisms for the biosynthesis of cellulose. It is difficult to envision a plausible mechanism for simultaneous synthesis and aggregation of antiparallel chains. It is perhaps for this reason that more recent proposals of parallel structures for native cellulose have been embraced by investigators of the mechanism of biosynthesis.

The contribution of spectroscopy to the early studies of structure was quite limited. An important contribution was made in the studies by Liang and Marchessault (24-26) wherein measurements of dichroism in infrared absorption of oriented specimens led to proposal of a particular hydrogen-bonding scheme. The differences between the spectra of celluloses I and II were explained in terms of differences in the packing of molecular chains and associated

variations in the hydrogen-bonding patterns. In another application, infrared absorption measurements were used as the basis of a crystallinity index by Nelson and O'Connor (27,28).

More recently, a number of new structure sensitive techniques have been developed, and they have been applied to studies of cellulose. These include Raman spectroscopy and Solid State ^{13}C Nuclear Magnetic Resonance, in the experimental arena, and conformational energy calculations in the theoretical domain. These are more recent contributions and are the subjects of subsequent sections in this chapter and later chapters in these proceedings.

Diffractometric Studies

As noted by Kakudo and Kasai, the primary difficulty in structural studies on polymeric fibers is that the number of reflections usually observed in diffractometric studies are quite limited. In the case of cellulose it is generally difficult to obtain more than 50 reflections. Consequently it becomes necessary to minimize the number of structural coordinates to be determined from the data by adopting plausible assumptions concerning the structure of the monomeric entity. The limited scattering data are then used to determine the orientation of the monomer units with respect to each other. In the majority of diffractometric studies of cellulose published so far, the monomeric entity has been chosen as the anhydroglucose unit. Thus, structural information from single crystals of glucose is implicitly incorporated in the analyses of the structure of cellulose. The coordinates which are adjusted in search of a fit to the diffractometric data include those of the primary alcohol group at C6, those of the glycosidic linkage, and those defining the positions of the chains relative to each other.

In addition to selection of the structure of the monomer as the basis for defining the internal coordinates of the repeat unit, the possible structures are usually further constrained by taking advantage of any symmetry possessed by the unit cell. The symmetry is derived from the systematic absence of reflections which are forbidden by the selection rules for a particular space group. In the case of cellulose, the simplification usually introduced is the application of the symmetry of space group $P2_1$, which includes a twofold screw axis parallel to the direction of the chains. The validity of this simplification remains the subject of controversy, however, because the reflections which are disallowed under the selection rules of the space group are in fact frequently observed. In most of the studies these reflections, which are usually weak relative to the other main reflections, are assumed to be negligible. The controversy continues because the relative intensities can be influenced by experimental conditions such as the periods of exposure of the diffractometric plates. Furthermore, the disallowed reflections tend to be more intense in electron diffractometric measurements than in x-ray diffraction measurements. Thus, more often than not, investigators using electron diffraction challenge the validity of the assumption of twofold screw axis symmetry.

The key assumption with respect to symmetry, however, is not the existence of the twofold screw axis as an element of the symmetry of the unit cell, but rather the additional assumption that

this axis coincides with the axis of the molecular chains of cellulose. This latter assumption has, implicit in it, a number of additional constraints on the possible structures which can be derived from the data. It requires that adjacent anhydroglucose units are related to each other by a rotation of 180 degrees about the axis, accompanied by a translation equivalent to half the length of the unit cell in that direction; it is implicit, therefore, that adjacent anhydroglucose units are symmetrically equivalent and, correspondingly, that alternating glycosidic linkages along the chain are symmetrically equivalent.

If the assumption concerning coincidence of the twofold screw axis and the molecular chain axis were excluded for example by locating the twofold screw axis between the molecular chains though still parallel to the chain axes, the diffractometric patterns would admit nonequivalence of alternate glycosidic linkages along the molecular chain, as well as the nonequivalence of adjacent anhydroglucose units. This possibility has been ignored, however, in large part because it requires expansion of the number of internal coordinates which have to be determined from the diffractometric data. Furthermore, it excludes the possibility of antiparallel alignment of chains in the unit cell.

The assumptions that the unit cell possesses the symmetry of space group $P2_1$ and that the twofold axis is coincident with the chain axis, do in fact meet a criterion long honored in scientific studies, namely, William of Ockham's principle of economy, which requires that the most simple hypothesis consistent with observations should always be adopted. Clearly the structure based on the anhydroglucose as the repeat unit is the most simple structure that accounts for the majority of the diffractometric data. Furthermore, the diffractometric data available are not sufficient to allow refinement of a structure possessing many more degrees of freedom, as would be the case if the twofold axis were not assumed coincident with the chain axis.

The assumptions concerning the symmetry of the unit cell noted above have been the basis of recent refinements of the structure of cellulose I. In one such refinement (17) the forbidden reflections were simply assumed negligible, and the intensity data from Valonia cellulose were used to arrive at a final structure. In another study, the inadequate informational content of the diffractometric data was complemented with analyses of lattice packing energies (29); the final structures were constrained to minimize the packing energy as well as optimizing the fit to the diffractometric data. Here the assumptions implicit in the weighting of the potential functions which are used in the energy calculations, further complicate the interpretations. As noted by French, et al. in a subsequent chapter in these proceedings, the structures derived in these two studies, though both based on parallel chain arrangements, are nevertheless very different crystal structures. When the same convention is applied in defining the axes of the crystal lattice, the structure most favored in one analysis is strongly rejected in the other. Furthermore, neither of these is strongly favored over yet a third, antiparallel structure (30).

The structures of oligomers are another important source of relevant information cited by Kakudo and Kasai. The implications of

the structures of the disaccharides have been considered by Atalla (31) and were the basis for reassessment of the second assumption concerning symmetry noted above. Structures with alternating nonequivalent glycosidic linkages were found more consistent with spectroscopic data (32).

Studies of oligomers have been extended in two chapters in the present volume, with the comparisons made primarily with structures proposed for cellulose II. Sakthivel, et al. applied the Rietveld crystal structure method to cellotetraose. Their results favor a parallel arrangement of chains in the unit cell, with individual chains possessing near twofold screw axis symmetry.

In a study of a number of oligomers, Henrissat, et al. used a multidisciplinary approach to examine the matter of the valid repeat unit. Their conformational analyses and ^{13}C NMR spectra were interpreted in terms of nonequivalent glycosidic linkages in the individual chains, but the diffraction data were found most consistent with an antiparallel structure.

Spectroscopy

Spectroscopic studies are useful in structural investigations because they provide information which is complementary to that derived from diffractometric data. The information derived from spectra is not directly related to the coordinates of molecules in the unit cell. The spectra are, however, sensitive to the values of internal coordinates which define molecular structure. Thus they provide a basis for testing the degrees of equivalence of structures. Very often also, specific spectral features can be identified with particular functional groups defined by distinctive sets of internal coordinates.

Two classes of spectral studies have been applied for the first time during the past decade as the basis of structural studies of cellulose. These are Raman spectroscopy, and solid state ^{13}C NMR using the CP/MAS technique. Both have raised questions concerning the assumptions about symmetry incorporated in the diffractometric studies. And while they cannot provide direct information concerning the structures, they establish criteria that any structure must meet to be regarded as an adequate model. The information from spectroscopic studies represents one of the major portions of the phenomenology that any acceptable structural model must rationalize.

Although the new spectral methods have also found application in investigations of structural changes induced by mechanical treatments or by treatments with swelling agents, the following discussion will be limited to studies which have focused on questions of structure. The results of such studies have to be rationalized by any model derived from crystallographic investigations and thus provide tests of consistency complementary to the diffractometric data, in the sense set forth by Kakudo and Kasai.

Raman Spectroscopy. Raman spectroscopy is the common alternative to infrared spectroscopy for investigating molecular vibrational states and vibrational spectra. It has enjoyed a significant revival since the development of laser sources for excitation of the spectra. Its key advantage in the present context is that it is

primarily sensitive to the skeletal vibrations of the cellulose molecule, with the mode of packing in the lattice having only secondary effects. This feature is a consequence of the dependence of Raman spectral activity of molecular vibrations on changes in the polarizability of vibrating bond systems, rather than changes in associated molecular dipoles. The most intense contributions to the spectra are due to bond systems which are predominantly covalent in character, with the more polar systems resulting in much weaker bands.

In the first detailed comparison of the Raman spectra of celluloses I and II, it was concluded that the differences between the spectra, particularly in the low frequency region, could not be accounted for in terms of chains possessing the same conformation but packed differently in the different lattices (33). As noted above, that had been the general interpretation of diffractometric studies of the two most common allomorphs. The studies of the Raman spectra led to the proposal that two different stable conformations of the cellulose chains occur in the different allomorphs.

In order to establish the differences between the conformations, information from other sources was considered. The results of published conformational energy calculations suggested two stable conformations for the glycosidic linkages (34,35). These represent relatively small left-handed and right-handed departures from the conformation of the glycosidic linkage in a twofold helical structure. They are well approximated, respectively, by the experimentally observed conformations of the glycosidic linkages in the crystal structures of the model disaccharides cellobiose (36) and methyl-β-cellobioside (37).

An analysis of the vibrational spectra in the OH stretching region for both the model disaccharides and for celluloses I and II suggested that nonequivalent glycosidic linkages alternate along the molecular chains (31). The solid state ^{13}C NMR spectra were found consistent with this model (38), although alternative interpretations are also possible. Finally the Raman spectra in the methylene bending region indicated that the C6 carbons occur in two nonequivalent environments in cellulose I but appear merged into a single set in cellulose II (39).

The results of the spectroscopic studies were interpreted in terms of nonequivalence of adjacent anhydroglucose units in the molecular chains, requiring the basic repeat unit of structure to be taken as the dimeric anhydrocellobiose unit. The difference between cellulose I and II was associated with the locus of the nonequivalence. In cellulose II it was thought to be at the glycosidic linkages, while in cellulose I it was taken to be centered at C6 and the adjacent segment of the pyranose rings.

To reconcile the conclusions outlined above with the requirements of chain packing, the proposal was made that cellulose chains possess alternate left-handed and right-handed glycosidic linkages in sequence along the chain axes. The left-handed and right-handed linkages were envisioned as representing relatively small departures of the dihedral angles from those prevailing for a twofold helix. The degree of departure from the parameters of a twofold helix was seen as somewhat greater for cellulose II than for cellulose I. The

model is discussed in somewhat more detail in the chapter by Wiley and Atalla, later in this volume.

Solid State ^{13}C NMR Spectra.

The second important spectroscopic method which has been applied in investigating the structure of cellulose during the past decade is high resolution ^{13}C NMR of the solid state based on the CP-MAS technique. In this technique, cross polarization (CP) is used to enhance the ^{13}C signal, high power proton decoupling to eliminate dipolar couplings with protons, and magic angle spinning (MAS) of the sample about a particular axis relative to the field to eliminate chemical shift anisotropy. Application of this method results in acquisition of spectra of sufficiently high resolution so that chemically equivalent carbons which occur in magnetically nonequivalent sites can be distinguished.

Though the technique has been used by a number of different investigators (38,40-43), we focus here on the studies by VanderHart and Atalla as representative of the structural questions addressed (44,45). Some resonance multiplicities for chemically equivalent carbons occur in the spectra of all the celluloses investigated.

The spectra of high crystallinity samples of cellulose II showed clear splittings of the resonances associated with C4 and C1. These have been interpreted as evidence of nonequivalent glycosidic linkages along the molecular chains (38), though it has also been suggested that the splittings may be evidence for nonequivalent chains in the unit cell (43). The latter argument leaves open the question as to why the resonances for carbons 2, 3, 5, and 6 do not display similar splittings.

Perhaps the most significant new information derived from the CP-MAS spectra is that relating to the native celluloses. The spectra reveal multiplicities that cannot be interpreted in terms of a unique unit cell, even though they arise from magnetically nonequivalent sites in crystalline domains. The narrow lines observed have relative intensities which are neither constant among the samples of different native celluloses, nor are they in the ratios of small whole numbers as would be expected if they arose from different sites within a relatively small unit cell. VanderHart and Atalla proposed that native celluloses are composites of two distinct crystalline forms (44,45).

Spectra of the two forms were resolved through linear combination of the spectra of native celluloses possessing the two forms in different proportions. The two types were designated celluloses I_α and I_β. The I_α form was found to be dominant in celluloses from lower plant forms and bacterial celluloses, while the I_β form was found dominant in celluloses from higher plants.

In studies of the Raman spectra of different native celluloses, Atalla (32) concluded that the two forms I_α and I_β consist of molecular chains which have the same molecular conformation. In the chapter by Wiley and Atalla in the present volume, evidence is presented to suggest that though the molecular conformations are the same, the hydrogen-bonding patterns differ in the two forms.

VanderHart and Atalla also present additional ^{13}C NMR CP-MAS experiments in a subsequent chapter. These provide strong evidence for the existence of the I_α and I_β forms in native celluloses,

particularly those from the lower plants and bacterial cellulose. They do raise, however, some questions about the earlier estimates of the amount of I_α form in the native celluloses from the higher plants.

In yet another application of the CP-MAS ^{13}C NMR spectroscopy in studies of the structure of celluloses, Horii, et al. have introduced correlations between the chemical shifts and dihedral angles as the basis of developing new structural information. In a subsequent chapter in these proceedings they provide an overview of their studies correlating the chemical shifts of specific carbons with the values of the dihedral angles about bonds involving those carbons. By examining the values of chemical shifts for monomeric and oligomeric compounds of known structures, they have developed correlations which may be applied in translating the spectral information in a manner that is complementary to the diffractometric studies.

Multidisciplinary Studies

In addition to the studies outlined above, with primary focus on diffractometry or on spectroscopy, there have been, recently, a number of studies which recognize at the outset the type of constraints summarized by Kakudo and Kasai, and which begin with an integrated approach to the investigation of structure. Perhaps the best illustration of this approach is the work of Henrissat and coworkers noted earlier and outlined in a later chapter, focusing on oligomers clearly related to the structure of cellulose II.

In the work of Henrisat, et al., the x-ray diffractometric data of Poppleton and Mathieson (46) on cellotetraose was complemented with structural data on other oligomers, with CP-MAS ^{13}C NMR spectroscopy, with conformational energy calculations, and with molecular orbital calculations to determine some of the favored conformations. The difficulty of the problem of the structures of cellulose is perhaps best illustrated by some of the remaining ambiguities cited in this study.

Yet another set of interdisciplinary studies are represented by the work of Hayashi and coworkers, wherein they attempt to shed light on the questions of reversibility, or lack thereof, in transformations between the allomorphs of cellulose and its derivatives. In addition to their diffractometric studies reported in prior publications, they add in their contribution to the present symposium analyses of the infrared spectra as well as analyses of the CP-MAS ^{13}C NMR spectra. Their thesis is not inconsistent with the proposals of Atalla and coworkers concerning differences between the conformations of celluloses I and II. However, Hayashi and coworkers go beyond this by proposing that the differences in conformation can be preserved in the course of heterogeneous derivatization reactions, and also in the process of generating the other allomorphs of cellulose, namely celluloses III and IV, from the two primary allomorphs I and II.

The adoption of multidisciplinary approaches in the effort to shed light on the complex questions of structures is likely to expand in the future. The proceedings of this symposium are clear evidence both for the need and the value of such approaches.

Future Directions

The studies reviewed briefly above place the problem of the structures of cellulose in a promising perspective. Until the development of the new spectroscopic methods, the crystallographic studies were undertaken with little additional information from other sources, with the exception of some of the conformational energy calculations. These are useful, but they are sensitive to the nature of the potential functions used in the calculations and particularly to the manner in which the different potential functions are weighted.

As noted earlier, the crystallographic studies have sought the most simple model structure consistent with observations. Clearly the structure based on the anhydroglucose as the repeat unit is the most simple structure that accounts for the majority of the diffractometric data. Furthermore, the data available did not provide a basis for introducing departures from the most simple model, nor suggestions for its revision.

The new information from spectroscopic studies sheds new light in two key areas. The first is related to the complexity of the structures of the native celluloses. The second is that of the relationship between the structures of celluloses I and II.

It has been known for some time that the more crystalline native celluloses from algae and from Acetobacter xylinum produce diffraction patterns that have many features in common with those of the crystalline celluloses from the higher plants, such as ramie, but that cannot be indexed as simply or on the basis of the same unit cell. The new information from the CP-MAS ^{13}C NMR spectra, together with that from the Raman spectra, suggests some bases for understanding these differences, and directions for further explorations.

The key conclusion that is relevant here is that the native celluloses are composites of more than one crystaline form, but that the difference between the two forms lies not in the molecular conformation but in the hydrogen bonding patterns. Thus, it is possible that the native celluloses have unit cells with very similar atomic coordinates for the heavy atoms, but with different coordinates for the hydrogens. The similarities in the heavy atom locations could account for the many comonalities in the diffraction patterns, while the differences in the coordinates of the hydrogen atoms could be responsible for the differences between the patterns. This would account for the greater incidence of nonallowed reflections in the electron diffraction patterns.

It is not clear that a polymeric system with a composite structure, such as the one proposed above, represents a tractable crystallographic problem. However, any new insights concerning the discrepancies between the proposed simple structures and the observations are important, for they may suggest departures in new directions for investigation. A very significant implication of the proposal suggested above, to explain the discrepancies between the diffraction patterns, is that the primary determining factor in the structure of the native celluloses may be the shape of the molecules rather than the hydrogen bonding pattern. The proposal clearly implies that more than one hydrogen bonding pattern is consistent

with the organization of the heavy atoms in the molecular skeleton in the unit cell. The proposal has a number of other implications for further investigation, discussion of which is beyond the scope of the present chapter.

With respect to the comparison between celluloses I and II, the spectral data leave little question that the molecular conformations are indeed different. The chapter by Wiley and Atalla sets forth some of the evidence based on Raman spectroscopy. The validity of the theoretical arguments developed in support of the hypothesis that two distinct conformations do indeed occur has been demonstrated through its application in studies of model compounds. The most comprehensive is a study of the vibrational spectra of the inositols (47), wherein spectra of seven of the isomers were investigated and the effects of conformational differences accounted for.

The hypothesis that conformational differences occur is also supported by the differences between the CP-MAS ^{13}C NMR spectra of celluloses I and II. It is indeed not likely that the differences in the chemical shifts of the different carbons and the differences in the degrees of splittings of the C1 and C4 resonances can be accounted for in terms of structures adhering strictly to the assumption that the twofold screw axes coincide with the axes of the molecular chains.

The data arising from both spectroscopic methods clearly point to the need to explore the degree to which the diffraction data can be accounted for in terms of structures wherein the anhydrocellobiose unit is assumed to be the basic repeat unit in the crystallographic structure. The spectroscopic studies and the conformational energy calculations suggest that the departures from equivalence of the two anhydroglucose units need not be very large ones. This may indeed be the reason why the disallowed reflections appear to be weak in the diffraction patterns. On the other hand, the spectroscopic evidence suggests that the nature of these minor departures from symmetric equivalence of adjacent anhydroglucose units may be the key to some of the anomalies encountered in the structural studies.

It is clear that the new information developed from spectroscopic and multidisciplinary studies provides a basis for initiating diffractometric studies with a different set of constraints than those used in the past. The refinements are likely to be more complex, but the expectation is that the structures thus derived will more closely approximate the molecular structure of cellulose. Such models may then provide more comprehensive rationalizations of the phenomenology of cellulose.

Literature Cited

1. Kakudo, M.; Kasai, N. X-ray diffraction by polymers, Elsevier, New York, 1972, p. 285.
2. Arnott, S. In Fiber Diffraction Methods; ACS Symposium Series No. 141, American Chemical Society, Washington, DC, 1980, p. 1.
3. Atkins, E. D. T. In Fiber Diffraction Methods, ACS Symposium Series No. 141, American Chemical Society, Washington, DC, 1980, p. 31.

4. Tadokoro, H. In Fiber Diffraction Methods, ACS Symposium Series No. 141, American Chemical Society, Washington, DC, 1980, p. 43.
5. Tadokoro, H. Structure of crystalline polymers, Wiley, New York, 1979, p. 6.
6. Purves, C. B. In Cellulose and Cellulose Derivatives, Pt. I; Ott, E.; Spurlin, H. M.; Graffline, M. W., Eds., Interscience, New York, 1954, p. 29.
7. Flory, P. J. Principles of polymer chemistry, Cornell University Press, Ithaca, New York, 1953, p. 3.
8. Hermans, P. H. Physics and chemistry of cellulose fibers, Elsevier, New York, 1949, p. 3.
9. Jones, D. W. In Cellulose and Cellulose Derivatives, Pt. IV; Bikales, N. M.; Segal, L.; Eds., Wiley-Interscience, New York, 1971, p. 117.
10. Ellefsen, O.; Tonessen, B. A. In Cellulose and Cellulose Derivatives, Pt. IV, Bikales, N. M.; Segal, L.; Eds., Wiley-Interscience, New York, 1971, p. 151.
11. Tonessen, B. A.; Ellefsen, O. In Cellulose and Cellulose Derivatives, Pt. IV, Bikales, N. M.; Segal, L.; Eds., Wiley-Interscience, New York, 1971, p. 265.
12. Preston, R. D. The physical biology of plant cell walls; Chapman and Hall, London, 1974.
13. Frey-Wyssling, A. The plant cell wall; Gebruder Borntrager, Berlin, 1976.
14. Howsmon, J. A.; Sisson, W. A. In Cellulose and Cellulose Derivatives, Pt. I; Ott, E.; Spurlin, H. M.; Graffline, M. W.; Eds., Interscience, New York, 1954, p. 231.
15. Petipas, T.; Oberlin, M.; Mering, J. J. Polym. Sci. C 1963, 2, 423.
16. Norman, M. Text. Res. J. 1963, 33, 711.
17. Gardner, K. H.; Blackwell, J. Biopolymers 1974, 13, 1975.
18. Hebert, J. J.; Muller, L. L. J. Appl. Polym. Sci. 1974, 18, 3373.
19. Ref. 9; p. 138.
20. Ref. 14; p. 237.
21. Meyer, K. H.; Mark, H. Z. Physik. Chem. 1929, B2, 115.
22. Meyer, K. H.; Misch, L. Helv. Chim. Acta 1937, 20, 232.
23. Pierce, F. T. Trans. Faraday Soc. 1946, 42, 545.
24. Liang, C. Y.; Marchessault, R. H. J. Polym. Sci. 1959, 37, 385.
25. Liang, C. Y.; Marchessault, R. H. J. Polym. Sci. 1959, 39, 269.
26. Marchessault, R. H.; Liang, C. Y. J. Polym. Sci. 1960, 43, 71.
27. Nelson, M. L.; O'Connor, R. T. J. Appl. Polym. Sci. 1964, 8, 1311.
28. Nelson, M. L.; O'Connor, R. T. J. Appl. Polym. Sci. 1964, 8, 1325.
29. Sarko, A.; Muggli, R. Macromol. 1974, 7, 486.
30. French, A. D. Carbohydrate Res. 1978, 61, 67.
31. Atalla, R. H. Advances in Chemistry Series 1979, 181, 55.
32. Atalla, R. H. In Structure, Function and Biosynthesis of Plant Cell Walls, Dugger, W. M.; Bartinicki-Garcia, S.; Eds.,

American Society of Plant Physiologists, Rockville, MD, 1984, p. 381.
33. Atalla, R. H. Appl. Polym. Symp. 1976, 28, 659.
34. Reese, D. A.; Skerrett, R. J. Carbohydrate Res. 1968, 7, 334.
35. Melberg, S.; Rasmussen, K. Carbohydrate Res. 1979, 71, 25.
36. Chu, S. S. C.; Jeffrey, G. A. Acta Crystallogr. 1968, B24, 830.
37. Ham, J. T.; Williams, D. G. Acta Crystallogr. 1970, B29, 1373.
38. Atalla, R. H.; Gast, J. C.; Sindorf, D. W.; Bartuska, V. J.; Maciel, G. E. JACS 1980, 102, 3249.
39. Atalla, R. H. Proceedings of the International Symposium on Wood and Pulping Chemistry, SPCI Rept. No 38, Stockholm 1981, Vol. 1, p. 57.
40. Earl, W. L.; VanderHart, D. L. JACS 1980, 102, 3251.
41. Earl, W. L.; VanderHart, D. L. Macromol. 1981, 14, 570.
42. Maciel, G. E.; Kolodziejski, W. L.; Bertran, M. S.; Dale, B. E. Macromol. 1982, 15, 686.
43. Fyfe, C. A.; Dudley, R. L.; Stephenson, P. J.; Deslandes, Y.; Hamer, G. K.; Marchessault, R. H. JACS 1983, 105, 2469.
44. Atalla, R. H.; VanderHart, D. L. Science 1984, 223, 283.
45. VanderHart, D. L.; Atalla, R. H. Macromol. 1984, 17, 1465.
46. Poppleton, B. J.; Mathieson, A. McL. Nature 1968, 219, 1946.
47. Williams, R. M.; Atalla, R. H. J. Phys. Chem. 1984, 88, 508.